APPLICATION OF GENOMIC AND PROTEOMIC PROFILING TO IDENTIFY POTENTIAL BIOMARKERS OF ALPHA-PARTICLE RADIATION EXPOSURE IN HUMAN LUNG EPITHELIAL CELLS

CHAUHAN V., HOWLAND M., O'HARA S., BEATON L., BURNS T., STOCKI T.J., WILKINS R.C.

Health Canada, Environmental and Radiation Health Sciences Directorate, Ottawa, Ontario, K1A-1C1, Vinita_chauhan@hc-sc.gc.ca

Of the radiation types, alpha- (α) particles are of particular interest as they are an environmental concern, predominately due to inhalation of radon and its daughter progeny. Furthermore, α-particle emitters like Americium-241, Plutonium-238 and Polonium-210 have been identified as possible isotopes to be used in radiological dispersal devices. Thus, the identification of potential biomarkers to α-particle radiation exposure would be useful for the development of field deployable bioassays which could be used for both human risk assessment and public health protection. This study was designed to examine subtle gene and protein responses following exposure of human epithelial cells to α-particle radiation. Exponentially growing human epithelial cells were exposed to α-particle radiation from Americium (^{241}Am) electroplated discs at doses ranging from 0-1.5Gy. Following exposure, cells were analyzed for gene and protein expression using "omics" scale technology. RNA was extracted from α-exposed cell cultures 4 h and 24 h after exposure. Microarray analysis was then used to determine transcript expression levels. Concurrently, two-dimensional gel electrophoresis coupled with mass spectrometry was used to analyze protein expression in cell lysates exposed to 1.5 Gy of α-particle radiation. Alterations in expression-level of protein spots between control- and exposed- treatment groups were analyzed statistically by the PDQuest software using Student's t-test (at the significance level of $p<0.05$). Mass spectrometry was used to determine the identity of protein-spots with significantly altered expression-levels. Four hours post-exposure, a total of 4 and 51 genes were differentially expressed (FDR<0.05) at 0.3 Gy and 0.9 Gy of α-particle radiation respectively. Twenty-four hours post-exposure, the medium and high doses of α-radiation caused statistically significant changes (FDR<0.05) in 58 and 590 genes respectively. Sixteen genes were identified that may be reliable α-responsive genes as they exhibited both time- and dose-dependant properties. Gene ontology analysis of these differentially expressed genes suggests that α-particle radiation induces cell cycle arrest, apoptosis and cell signalling. Comparison of the 2-dimensional gel images between the control and α-particle exposed groups showed 15 upregulated protein spots and 1 downregulated protein spot, of which 4 were identified by mass spectrometry. These four proteins were induced by >2 fold and included Annexin A2 a cell motility protein, peroxiredoxin-6 a protein with protective properties against oxidative injury, proteasome

subunit α-type-3, a mediator of CDKN1A degradation and lastly HSPA8, a chaperone molecule involved in correct protein folding. Despite the need for further validation, the data suggest that alterations in expression-levels of specific genes/proteins may be unique indicators of α-radiation-exposure. These targets, therefore, may be potential biomarkers of α-particle radiation-exposure and could be used further for the development of fast and reliable bioassays.